

CLAIMS

What is claimed is:

1. A method of selectively detecting COX-2 activity in a sample, comprising:
 - a. adding a COX-2 selective substrate to the sample; and
 - b. detecting a metabolite of the COX-2 selective substrate, thereby indicating the COX-2 activity.
2. A method of measuring COX-1 activity in a sample, comprising:
 - a. adding a nonselective COX substrate and a COX-2 selective substrate to the sample;
 - b. allowing a period of time to pass;
 - c. measuring a first amount of a metabolite of the COX-1 substrate in the sample and a second amount of a metabolite of the COX-2 selective substrate metabolite; and
 - d. comparing the first amount and the second amount.
3. A method of detecting an activity of a COX-2 enzyme in a sample, comprising: detecting a PGH₂-EA metabolite in the sample, wherein the presence of the PGH₂-EA metabolite in the sample indicates the activity of the COX-2 enzyme.
4. A method of measuring an activity of a COX-2 enzyme in a sample, comprising:
 - a. quantifying an amount of a PGH₂-EA metabolite in the sample; and
 - b. relating the amount of the PGH₂-EA metabolite to the activity of the COX-2 enzyme.
5. A method of distinguishing a COX-2 activity from a COX-1 activity in a subject, comprising:
 - a. administering a COX-1 substrate and COX-2 selective substrate to the subject;
 - b. allowing a period of time to pass;
 - c. obtaining a sample from the subject;

- d. determining a first amount of a metabolite of the COX-1 substrate and a second amount of a metabolite of the COX-2 selective substrate; and
 - e. comparing the first amount to the second amount.
- 5 6. A method of detecting an activity of a COX-2 enzyme in a subject, comprising:
- a. obtaining a sample of the subject; and
 - b. detecting a PGH_2 -EA metabolite in the sample, wherein the presence of the PGH_2 -EA metabolite in the sample indicates the activity of the COX-2 enzyme in the subject.
- 10 7. The method of Claim 6, wherein the PGH_2 -EA metabolite is selected from the group consisting of PGB_2 -EA, PGD_2 -EA, PGE_2 -EA, $\text{PGF}_{2\alpha}$ -EA, TxB_2 -EA, 6-keto- $\text{PGF}_{1\alpha}$ -EA, 15-keto- PGE_2 -EA, 13,14-dihydro-15-keto- PGE_2 -EA, PGG_2 -EA, PGH_2 -EA, PGA_2 -EA, PGJ_2 -EA, PGJ_2 -EA derivatives, bicyclo- PGE_2 -EA, 6-keto- $\text{PGF}_{1\alpha}$ -EA, TxA_2 -EA and PGI_2 -EA.
- 15 8. The method of Claim 6, wherein the subject is a mammal.
9. The method of Claim 6, wherein the sample is urine.
10. The method of Claim 6, wherein the sample is selected from a group consisting of: blood, plasma, cerebrospinal fluid, saliva, sputum, bile, joint fluid, biopsy, and conditioned media from a cell culture.
- 20 11. The method of Claim 6, wherein the detecting step further comprises generating a mass chromatogram of the PGH_2 -EA metabolites.
12. The method of Claim 6, wherein the detecting step includes an immunoassay step.
- 25 13. A method of measuring an activity of a COX-2 enzyme in a subject, comprising:
- a. obtaining a sample of the subject;
 - b. measuring an amount of a PGH_2 -EA metabolite in the sample; and
 - c. relating the amount measured to the activity of the COX-2 enzyme.

14. The method of Claim 13, wherein the relating step further comprises comparing the amount measured to a standard value.
15. The method of Claim 13, wherein the relating step further comprises generating a standard curve.
- 5 16. The method of Claim 13, wherein the PGH₂-EA metabolite is selected from the group consisting of PGB₂-EA, PGD₂-EA, PGE₂-EA, PGF₂α-EA, TxB₂-EA, 6-keto-PGF₁α-EA, 15-keto-PGE₂-EA, 13,14-dihydro-15-keto-PGE₂-EA, PGG₂-EA, PGH₂-EA, PGA₂-EA, PGJ₂-EA, PGJ₂-EA derivatives, bicyclo-PGE₂-EA, 6-keto-PGF₁α-EA, TxA₂-EA and PGI₂-EA.
17. The method of claim 13, wherein the subject is a mammal.
18. The method of claim 13, wherein the sample is urine.
19. The method of claim 13, wherein the sample is selected from a group consisting of: blood, plasma, cerebrospinal fluid, saliva, sputum, bile, joint fluid, biopsy, and conditioned media from a cell culture.
20. The method of Claim 13, wherein the detecting step further comprises generating a mass chromatogram of the PGH₂-EA metabolites.
21. The method of Claim 13, wherein the detecting step includes an immunoassay step.
22. A method of screening for a tumor in a subject in need thereof, comprising:
- 20 a. obtaining a sample of the subject; and
- b. detecting a PGH₂-EA metabolite in the sample; wherein the presence of the PGH₂-EA metabolite is indicative of the tumor in the subject.
23. A method of screening for a tumor in a subject in need thereof, comprising:
- a. obtaining a sample of the subject;
- 25 b. measuring an amount of a PGH₂-EA metabolite in the sample; and
- c. relating the amount measured to an existence of the tumor.
24. A method of monitoring an anticancer treatment, comprising:

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- a. obtaining a first sample of a patient;
 - b. measuring a first amount of PGH_2 -EA metabolite in the first sample;
 - c. obtaining a second sample from the patient after the patient undergoes anticancer therapy;
 - d. measuring a second amount of the PGH_2 -EA metabolite in the second sample; and
 - e. determining a change in the second amount relative to the first amount, wherein the change determined is indicative of the effectiveness of the anticancer treatment.
- 10 25. A method of detecting an inflammation in a subject in need thereof, comprising:
- a. obtaining a sample of the subject; and
 - b. detecting an amount of a PGH_2 -EA metabolite in the sample, wherein an inflammation is indicated when the amount detected equals or exceeds a threshold value.
- 15 26. A method of measuring an inflammation in a subject in need thereof, comprising:
- a. obtaining a sample of the subject; and
 - b. detecting an amount of a PGH_2 -EA metabolite in the sample, wherein an inflammation is indicated when the amount measured equals or exceeds a
- 20 threshold value.
- 25 27. A method of monitoring an anti-inflammation therapy in a subject in need thereof, comprising:
- a. obtaining a first sample from the subject;
 - b. measuring a first amount of a PGH_2 -EA metabolite in the first sample;
 - c. obtaining a second sample from the patient after the anti-inflammation therapy;

- d. measuring a second amount of the COX-2 specific metabolite in the second sample; and
- e. determining a change in the second amount relative to the first amount, wherein the change determined is indicative of the effectiveness of the anti-inflammation therapy.

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- 28. A composition comprising: a label for detecting a PGH_2 -EA metabolite.
- 29. The composition of Claim 28, further comprising an isolated PGH_2 -EA metabolite including an isotopic label.
- 30. The method of Claim 28, wherein the PGH_2 -EA metabolite is selected from the group consisting of PGB_2 -EA, PGD_2 -EA, PGE_2 -EA, $\text{PGF}_{2\alpha}$ -EA, TxB_2 -EA, 6-keto- $\text{PGF}_{1\alpha}$ -EA, 15-keto- PGE_2 -EA, 13,14-dihydro-15-keto- PGE_2 -EA, PGG_2 -EA, PGH_2 -EA, PGA_2 -EA, PGJ_2 -EA, PGJ_2 -EA derivatives, bicyclo- PGE_2 -EA, 6-keto- $\text{PGF}_{1\alpha}$ -EA, TxA_2 -EA and PGI_2 -EA.
- 31. The composition of Claim 28, further comprising an isolated PGH_2 -EA metabolite including a non-positron emitting isotopic label.
- 32. The composition of Claim 28, further comprising an isolated PGH_2 -EA metabolite including an isotopic label selected from the group consisting of ^2H , ^3H , ^{13}C , and ^{14}C .
- 33. The composition of Claim 28, further comprising an PGH_2 -EA metabolite including a fluorescent label.
- 34. A process for making an isolated PGH_2 -EA metabolite including a label comprising: reacting a COX-2 metabolite with a labeled ethanolamide.
- 35. The process of Claim 34, wherein the label is isotopic.
- 36. The process of Claim 34, wherein the label is nonpositron emitting.
- 37. The process of Claim 34, wherein the label is selected from the group consisting of ^2H , ^3H , ^{13}C , and ^{14}C .
- 38. The process of Claim 34, wherein the label is fluorescent.

39. A process for making an isolated PGH₂-EA metabolite including a label comprising: reacting a labeled COX-2 metabolite with ethanolamide.
40. The process of Claim 39, wherein the label is isotopic.
41. The process of Claim 39, wherein the label is nonpositron emitting.
- 5 42. The process of Claim 39, wherein the label is selected from the group consisting of ²H, ³H, ¹³C, and ¹⁴C.
43. The process of Claim 39, wherein the label is fluorescent.
44. An article of manufacture comprising, packaged together:
- a. a vessel containing an isolated antibody against a PGH₂-EA metabolite; and
 - b. a set of instructions delineating a process of measuring a COX-2 specific activity.
45. An article of manufacture comprising, packaged together:
- a. a vessel containing at least one labeled PGH₂-EA metabolite; and
 - b. a set of instructions delineating a process of measuring a COX-2 specific activity.
46. An antibody that binds specifically to a COX-2 metabolite ethanolamide.
47. An antibody that binds specifically to a PGH₂-EA metabolite.
48. An antibody that binds specifically to a PGE₂ ethanolamide.
- 20 49. A process of making an antibody that binds specifically to PGH₂-EA metabolites from a prostaglandin with substituted cyclopentyl and amide moieties, comprising:
- a. protecting the cyclopentyl substituents and ethanolamide moiety of the prostaglandin to produce a protected PG-EA;
 - 25 b. chemically modifying the protected PG-EA with an appropriate conjugate to produce a protected, conjugated PG-EA;
 - c. deprotecting the conjugated PG-EA to generate an immunogen; and

d. purifying the immunogen.

50. A method of measuring an activity of a COX-2 enzyme in a subject, comprising:

a. administering an amount of a AEA to the subject;

b. obtaining a sample of the subject;

5 c. measuring an amount of a PGH₂-EA metabolites in the sample; and

d. relating the amount of the PGH₂-EA metabolites to the activity of the COX-2 enzyme.

51. The method of Claim 50, wherein the AEA includes a label.

52. The method of Claim 50, further comprising the step of comparing the amount measured to a standard.

53. A composition comprising: a prostaglandin D₂-ethanolamide and pharmaceutically acceptable salts thereof.

54. A composition comprising: a 6-keto-prostaglandin F_{1α}-ethanolamide and pharmaceutically acceptable salts thereof.